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The current study investigates whether polymorphisms in the CYP17 gene, which plays roles in estrogen biosynthesis, are associated with an altered risk of breast cancer in a population-based sample of women (400 cases and 400 controls) who participated in the Long Island Breast Cancer Study Project (LIBSCP). In addition, we are also exploring whether the effects of reproductive risk factors and exposure to exogenous estrogen are modified by CYP17 polymorphisms. In the past year, laboratory assays on CYP17 polymorphisms are being conducted in the Division of Environmental Health Sciences of Columbia University. The polymorphisms are being determined by PCR-RFLP methods with a digestion by appropriate restriction enzymes. To date, assays on 300 cases and 300 controls have been performed. Delays in subject and sample selection in the parent LIBSCP study delayed Task 1, thus delaying the start of laboratory analyses. Therefore, Task 2 will not be completed until the end of September 1999. Entry of laboratory data to computers and merging of laboratory data with questionnaire data (Task 3) are estimated to begin in October 1999. Data analysis, manuscript preparation and report writing (Tasks 4 & 5) will take place in the coming year.

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Cytochrome P450-17alpha Polymorphism and Risk of Breast Cancer
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Introduction

Cumulative exposure of circulating estrogen to the breast is linked to the etiology of breast cancer with several studies[1, 2] suggesting that circulating estradiol (the active form of estrogen) is significantly elevated in breast cancer patients compared to controls in both high and low risk populations[3-6]. This variation in circulating estradiol may be due in part to genetic polymorphisms affecting the activity of the enzymes responsible for the metabolism and cellular binding of estrogen in individuals. The enzyme, whose polymorphisms have demonstrated the most potential in the etiology of breast cancer, is the cytochrome P450c17α (CYP17) enzyme. Therefore, the current investigation is investigating whether polymorphisms of the CYP17 gene are associated with an altered risk of breast cancer in a population-based sample of women (400 controls and 400 cases) who participated in the Long Island Breast Cancer Study Project (LIBCSP). Further, we are also exploring whether the effects of reproductive risk factors (e.g., age at menarche, menopause, last child birth and parity) and exposure to exogenous estrogen like hormone replacement therapy, oral contraceptives or estrogen-mimicking substances such as organochlorine pesticides on breast cancer are modified by CYP17 polymorphisms.

Body

Subject and sample selection was delayed in the parent study (LIBSCP) and thus Task I which involved randomly selecting 400 cases and 400 controls (frequency matched to cases by five-year age groups) in batches, using code numbers from among women who had participated in the LIBCSP, was not completed until Month 4. These women provided blood samples and completed a questionnaire for the LIBSCP. Approximately 75% of study participants are postmenopausal in this randomly selected subgroup of the LIBCSP. As expected, the majority of cases and controls randomly selected for this study reflected the racial make-up of the LIBSCP population which is 90% Caucasian, 7% Black and 3% other races.

All laboratory analyses are being done in the laboratory of Dr. Regina Santella in the Division of Environmental Health Sciences of Columbia University. Genomic DNA was isolated from all cases and controls using standard phenol/chloroform extraction method. Polymorphism in the CYP17 gene is being determined by PCR-RFLP methods with the use of appropriate restriction enzymes. Briefly, genomic DNA was amplified by PCR using CYP17-specific primers. Following PCR amplification, digestion of amplified DNA was accomplished by MspI A1 restriction enzyme. Restriction Fragment Length Polymorphism (RFLP) patterns are being examined by agarose gel electrophoresis after staining with ethidium bromide to detect CYP17 A1 and A2 alleles. The polymorphisms are being identified as follows: 1) a base change in both alleles (A2/A2 genotype) identified if two bands of DNA on the gel are generated, 2) heterozygous (A1/A2) individuals are identified as having three bands on the gel since the enzyme cuts only the mutated allele and not the wild type allele, and 3) individuals who are homzygous (A1/A1) for the alleles produce one band of the uncut PCR product on gel. CYP17 assays completed to date were performed and interpreted blinded to subject characteristics and case-control status. To date these laboratory assays have been performed on only 300 cases and 300 controls as a result of the delay in beginning Task I, as previously mentioned. However, laboratory assays for the remaining cases and controls are expected to be completed by the end of September 1999. Task 3, which includes entry of laboratory data to computers and merging of laboratory data with questionnaire data will be completed in October/November 1999.

Data analysis (Task 4) as well as manuscript preparation and report writing (Task 5) will commence in the coming year.

Key Research Accomplishments
Not applicable at this time.

Reportable Outcomes
Not applicable at this time.

Conclusions
Not applicable at this time.

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